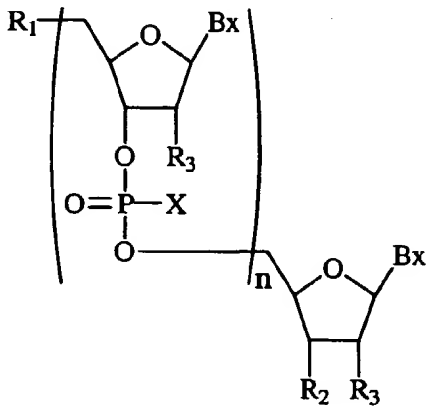
Renumbered Claim Set

This listing of claims will replace all prior versions, and listings, of claims in the application.

Listing of Claims:

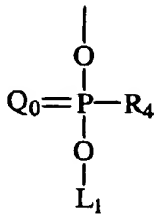
1-21. (canceled)

22. (currently amended) A process for preparing an oligonucleotide having the formula:



wherein:

R_1 is a group having the formula:

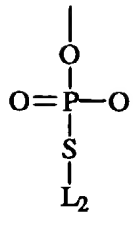


;

Q_0 is O or S;

R_4 is O^-, hydroxyl, or a protected hydroxyl;

R_2 is hydroxyl, a protected hydroxyl or a group having the formula:



;

each R_3 is H, a 2'-substituent group or a protected 2'-substituent group;

each X is, independently, O^-, hydroxyl, protected hydroxyl, or -S-L_3;

each Bx is an optionally protected heterocyclic base moiety;

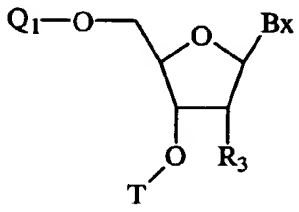
n is from 3 to about 50; and

L_1, L_2 and each of said L_3 are, independently, a cholesterol, phospholipid, biotin, phenazine, phenanthridine, anthraquinone, acridine, fluorescein, rhodamine, or coumarin~~, or dye;~~

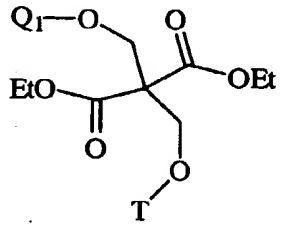
wherein said R_1 and at least one of said R_2 or said X comprise a cholesterol, phospholipid, biotin, phenazine, phenanthridine, anthraquinone, acridine, fluorescein, rhodamine, or coumarin;

comprising the steps of:

a) providing a derivatized solid support for oligonucleotide synthesis, said derivatized solid support being derivatized with a group having one of the structures:



or



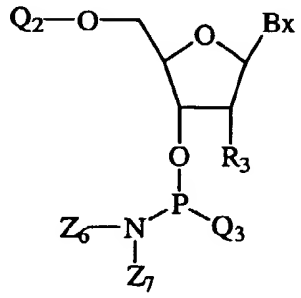
wherein

T is a bifunctional linking moiety linked to the solid support; and

Q_1 is an acid labile hydroxyl protecting group;

b) treating said derivatized solid support with an acidic reagent to deblock said acid labile hydroxyl protecting group to give a free hydroxyl group;

c) reacting said free hydroxyl group with a phosphoramidite composition to form an extended compound, said phosphoramidite composition having the formula:



wherein

Q_2 is a 5'-terminal acid labile hydroxyl protecting group;

Q_3 is a phosphorus protecting group; and

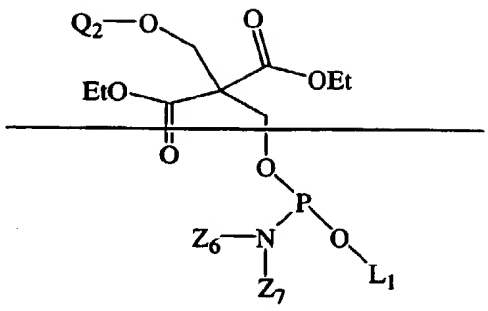
Z_6 and Z_7 are, independently, C_{1-6} alkyl;

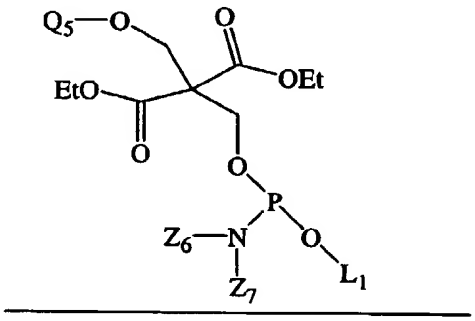
or Z_6 and Z_7 are joined together to form a 4- to 7-membered heterocyclic ring system including the nitrogen atom to which Z_6 and Z_7 are attached, wherein said ring system optionally includes at least one additional heteroatom selected from O, N and S;

d) oxidizing said extended compound to form an oxidized compound, or treating said extended compound with an acidic reagent to deblock said 5'-terminal acid labile hydroxyl protecting group of said extended compound to give a free hydroxyl group and repeating step c) at least one time followed by oxidizing said extended compound to form an oxidized compound;

e) treating said oxidized compound with an acidic reagent to deblock said acid labile hydroxyl protecting group to give a free hydroxyl group and repeating steps c) and d) at least three times to form an extended oxidized compound;

f) treating said extended oxidized compound with a reagent effective to deblock said protected hydroxyl group to give a free hydroxyl group and reacting said free hydroxyl group with a compound of formula:





thereby forming a 5'-functionalized compound;

wherein

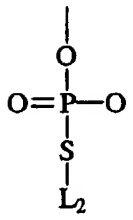
Q_5 is an acid labile hydroxyl protecting group;

g) treating said 5'-functionalized compound for a time and under conditions effective to remove at least one phosphorus protecting group giving at least one deblocked phosphorothioate linkage; and

h) reacting said deblocked phosphorothioate linkage with a cholesterol, phospholipid, biotin, phenazine, phenanthridine, anthraquinone, acridine, fluorescein, rhodamine, or coumarin, that is reactive with and forms a covalent bond with said deblocked phosphorothioate linkage to give said oligonucleotide.

23. (original) The process of Claim 22 further comprising the step of treating said 5'-functionalized compound with a capping agent to form a capped compound.

24. (original) The process of Claim 22 wherein said R_2 is a group having the formula:



25. (original) The process of Claim 24 wherein L_1 is different from L_2.

26. (original) The process of Claim 22 wherein at least one of said X is -S-L_3.

27. (original) The process of Claim 26 wherein L_1 is different from L_3.

28. (canceled)

29. (canceled)

30. (previously presented) The process of Claim 22 wherein each of said Q_3 is independently selected from the group consisting of cyanoethyl, diphenylsilylethyl, cyanobutenyl, cyano *p*-xylyl (CPX), methyl-N-trifluoroacetyl ethyl (META) and acetoxy phenoxy ethyl (APOE) groups.

31. (original) The process of Claim 22 wherein said 5'-functionalized compound is treated in step g) to remove all phosphorus protecting groups.

32. (original) The process of Claim 22 wherein n is from about 8 to about 30.

33. (original) The process of Claim 32 wherein n is from about 15 to about 25.

34. (original) The process of Claim 22 wherein each of said Q_1 and Q_2 is independently selected from the group consisting of trimethoxytrityl, dimethoxytrityl (DMT),

monomethoxytrityl, 9-phenylxanthen-9-yl (Pixyl) and 9-(p-methoxyphenyl)xanthen-9-yl (Mox).

12 ~~35.~~ (original) The process of Claim ~~22~~ 1 wherein each of said Bx is independently selected from the group consisting of adenine, guanine, thymine, cytosine, uracil, 5-methylcytosine (5-me-C), 5-hydroxymethyl cytosine, xanthine, hypoxanthine, 2-aminoadenine, alkyl derivatives of adenine and guanine, 2-thiouracil, 2-thiothymine, 2-thiocytosine, 5-halouracil, 5-halocytosine, 5-propynyl uracil, 5-propynyl cytosine, 6-azo uracil, 6-azo cytosine, 6-azo thymine, 5-uracil (pseudouracil), 4-thiouracil, 8-substituted adenines and guanines, 5-substituted uracils and cytosines, 7-methylguanine, 7-methyladenine, 8-azaguanine, 8-azaadenine, 7-deazaguanine, 7-deazaadenine, 3-deazaguanine and 3-deazaadenine.

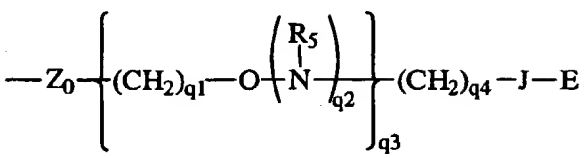
13 ~~36.~~ (original) The process of Claim ~~22~~ 1 wherein at least one of said L_1, L_2, and L_3 is attached to the oligonucleotide through a linking group.

14 ~~37.~~ (original) The process of Claim ~~36~~ 13 wherein the linking group comprises a dialkylglycerol linker.

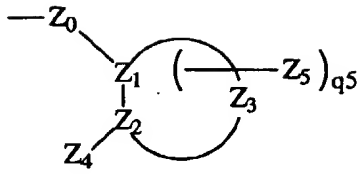
15 ~~38.~~ (original) The process of Claim ~~22~~ 1 wherein each of said Z_6 and Z_7 is isopropyl.

16 ~~39.~~ (original) The process of Claim ~~22~~ 1 wherein each R_3 is, independently, C_1-C_{20} alkyl, C_2-C_{20} alkenyl, C_2-C_{20} alkynyl, C_5-C_{20} aryl, O-alkyl, O-alkenyl, O-alkynyl, O-alkylamino, O-alkylalkoxy, O-alkylaminoalkyl, O-alkyl imidazole, thiol, S-alkyl, S-alkenyl, S-alkynyl, NH-alkyl, NH-alkenyl, NH-alkynyl, N-dialkyl, O-aryl, S-aryl, NH-aryl, O-aralkyl, S-aralkyl, NH-aralkyl, N-phthalimido, halogen keto, carboxyl, nitro, nitroso, nitrile, trifluoromethyl, trifluoromethoxy, imidazole, azido, hydrazino, hydroxylamino, isocyanato, sulfoxide, sulfone, sulfide, disulfide, silyl, heterocycle, carbocycle, polyamine, polyamide, polyalkylene glycol, and polyether;

or each substituent group has one of formula I or II:



I



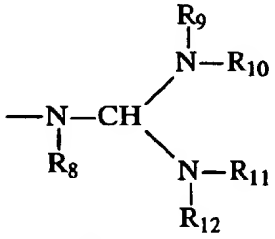
II

wherein:

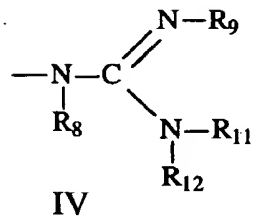
Z_0 is O, S or NH;

J is a single bond, O or C(=O);

E is C_1-C_{10} alkyl, $N(R_5)(R_6)$, $N(R_5)(R_7)$, $N=C(R_5)(R_6)$, $N=C(R_5)(R_7)$ or has one of formula III or IV;



III



IV

each R_8, R_9, R_{10}, R_{11} and R_{12} is, independently, hydrogen, $C(O)R_{13}$, substituted or unsubstituted C_1-C_{10} alkyl, substituted or unsubstituted C_2-C_{10} alkenyl, substituted or unsubstituted C_2-C_{10} alkynyl, alkylsulfonyl, arylsulfonyl, a chemical functional group or a conjugate group, wherein the substituent groups are selected from hydroxyl, amino, alkoxy, carboxy, benzyl, phenyl, nitro, thiol, thioalkoxy, halogen, alkyl, aryl, alkenyl and alkynyl;

or optionally, R_9 and R_{10}, together form a phthalimido moiety with the nitrogen atom to which they are attached;

or optionally, R_{11} and R_{12}, together form a phthalimido moiety with the nitrogen atom to which they are attached;

each R_{13} is, independently, substituted or unsubstituted C_1-C_{10} alkyl, trifluoromethyl, cyanoethyloxy, methoxy, ethoxy, t-butoxy, allyloxy, 9-fluorenylmethoxy, 2-(trimethylsilyl)-ethoxy, 2,2,2-trichloroethoxy, benzyloxy, butyryl, iso-butyryl, phenyl or aryl;

R_5 is T-L,

T is a bond or a linking moiety;

L is a chemical functional group, a conjugate group or a solid support material;

each R_5 and R_6 is, independently, H, a nitrogen protecting group, substituted or unsubstituted C_1-C_{10} alkyl, substituted or unsubstituted C_2-C_{10} alkenyl, substituted or unsubstituted C_2-C_{10} alkynyl, wherein said substitution is OR_3, SR_3, NH_3^+, $N(R_{14})(R_{15})$, guanidino or acyl where said acyl is an acid amide or an ester;

or R_5 and R_6, together, are a nitrogen protecting group or are joined in a ring structure that optionally includes an additional heteroatom selected from N and O;

or R_{21}, T and L, together, are a chemical functional group;

each R_{14} and R_{15} is, independently, H, C_1-C_{10} alkyl, a nitrogen protecting group, or R_{14} and R_{15}, together, are a nitrogen protecting group;

or R_{14} and R_{15} are joined in a ring structure that optionally includes an additional heteroatom selected from N and O;

Z_4 is OX, SX, or $N(X)_2$;

each X is, independently, H, C_1-C_8 alkyl, C_1-C_8 haloalkyl, $C(=NH)N(H)R_{16}$, $C(=O)N(H)R_{16}$ or $OC(=O)N(H)R_{16}$;

R_{16} is H or C_1-C_8 alkyl;

Z_1, Z_2 and Z_3 comprise a ring system having from about 4 to about 7 carbon atoms or having from about 3 to about 6 carbon atoms and 1 or 2 heteroatoms wherein said heteroatoms are selected from oxygen, nitrogen and sulfur and wherein said ring system is aliphatic, unsaturated aliphatic, aromatic, or saturated or unsaturated heterocyclic;

Z_5 is alkyl or haloalkyl having 1 to about 10 carbon atoms, alkenyl having 2 to about 10 carbon atoms, alkynyl having 2 to about 10 carbon atoms, aryl having 6 to about 14 carbon atoms, $N(R_5)(R_6)$ OR_5, halo, SR_5 or CN;

each q_1 is, independently, an integer from 1 to 10;

each q_2 is, independently, 0 or 1;

q_3 is 0 or an integer from 1 to 10;

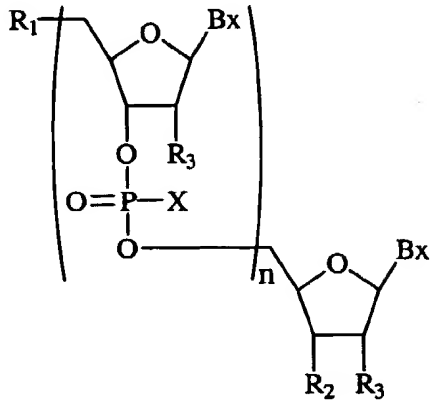
q_4 is an integer from 1 to 10;

q_5 is from 0, 1 or 2; and

provided that when q_3 is 0, q_4 is greater than 1.

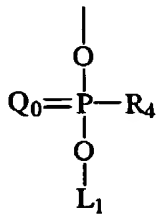
40-49. (canceled)

17 50. (currently amended) A process for preparing an oligonucleotide having the formula:



wherein:

R_1 is a group having the formula:

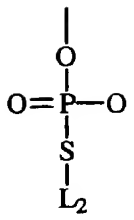


;

Q_0 is O or S;

R_4 is O^-, hydroxyl, or a protected hydroxyl;

R_2 is hydroxyl, a protected hydroxyl or a group having the formula:



;

each R_3 is H, a 2'-substituent group or a protected 2'-substituent group;

each X is, independently, O^-, hydroxyl, a protected hydroxyl, or -S-L_3;

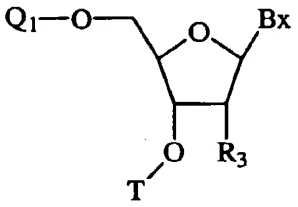
each Bx is an optionally protected heterocyclic base moiety;

n is from 3 to about 50; and

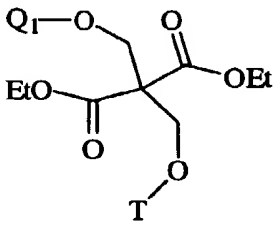
L_1, L_2 and each of said L_3 are, independently, a cholesterol, phospholipid, biotin, phenazine, phenanthridine, anthraquinone, acridine, fluorescein, rhodamine, or coumarin;

comprising the steps of:

a) providing a derivatized solid support for oligonucleotide synthesis, said derivatized solid support being derivatized with a group having one of the structures:



or



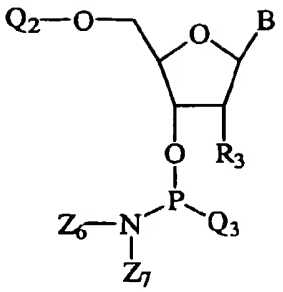
wherein

T is a bifunctional linking moiety linked to the solid support; and

Q_1 is an acid labile hydroxyl protecting group;

b) treating said derivatized solid support with an acidic reagent to deblock said acid labile hydroxyl protecting group to give a free hydroxyl group;

c) reacting said free hydroxyl group with a phosphoramidite composition to form an extended compound, said phosphoramidite composition having the formula:



wherein

Q_2 is a 5'-terminal acid labile hydroxyl protecting group;

Q_3 is a phosphorus protecting group; and

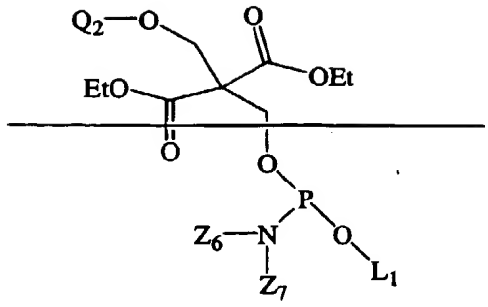
Z_6 and Z_7 are, independently, C_{1-6} alkyl;

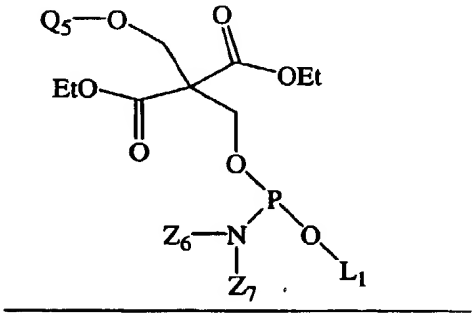
or Z_6 and Z_7 are joined together to form a 4- to 7-membered heterocyclic ring system including the nitrogen atom to which Z_6 and Z_7 are attached, wherein said ring system optionally includes at least one additional heteroatom selected from O, N and S;

d) oxidizing said extended compound to form an oxidized compound, or treating said extended compound with an acidic reagent to deblock said 5'-terminal acid labile hydroxyl protecting group of said extended compound to give a free hydroxyl group and repeating step c) at least one time followed by oxidizing said extended compound to form an oxidized compound;

e) treating said oxidized compound with an acidic reagent to deblock said acid labile hydroxyl protecting group to give a free hydroxyl group and repeating steps c) and d) at least three times to form an extended oxidized compound;

f) treating said extended oxidized compound with an acidic reagent effective to deblock said 5'-terminal acid labile hydroxyl protecting group to give a free hydroxyl group and reacting said free hydroxyl group with a compound of the formula:





thereby forming a 5'-functionalized compound;

wherein

Q_5 is an acid labile hydroxyl protecting group;.

18 51. (original) The process of Claim 17 further comprising the step of treating said 5'-functionalized compound with a capping agent to form a capped compound.

19 52. (original) The process of Claim 17 wherein at least one of said L_1, L_2, and L_3 is attached to the oligonucleotide through a linking group.

20 53. (original) The process of Claim 19 wherein the linking group comprises a dialkylglycerol linker.

21 54. (original) The process of Claim 17 wherein each of said Z_6 and Z_7 is isopropyl.

55. (canceled)

56. (canceled)

22 57. (original) The process of Claim ~~50~~ 17 wherein L_1 is different from L_2 and L_3.

23 58. (original) The process of Claim ~~50~~ 17 wherein each of said Q_3 is independently selected from the group consisting of cyanoethyl, diphenylsilylethyl, cyanobutenyl, cyano *p*-xylyl (CPX), methyl-N-trifluoroacetyl ethyl (META) and acetoxy phenoxy ethyl (APOE) groups.

24 59. (original) The process of Claim ~~50~~ 17 wherein each of said Q_1 and Q_2 is independently selected from the group consisting of trimethoxytrityl, dimethoxytrityl (DMT), monomethoxytrityl, 9-phenylxanthen-9-yl (Pixyl) and 9-(p-methoxyphenyl)xanthen-9-yl (Mox).

25 60. (original) The process of Claim ~~50~~ 17 wherein each of said Bx is independently selected from the group consisting of adenine, guanine, thymine, cytosine, uracil, 5-methylcytosine (5-me-C), 5-hydroxymethyl cytosine, xanthine, hypoxanthine, 2-aminoadenine, alkyl derivatives of adenine and guanine, 2-thiouracil, 2-thiothymine, 2-thiocytosine, 5-halouracil, 5-halocytosine, 5-propynyl uracil, 5-propynyl cytosine, 6-azo uracil, 6-azo cytosine, 6-azo thymine, 5-uracil (pseudouracil), 4-thiouracil, 8-substituted adenines and guanines, 5-substituted uracils and cytosines, 7-methylguanine, 7-methyladenine, 8-azaguanine, 8-azaadenine, 7-deazaguanine, 7-deazaadenine, 3-deazaguanine and 3-deazaadenine.